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Tissue Polypeptide-Specific Antigen in Pediatric Patients: Assessment of Normal Values

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Measurement of serum concentrations of tissue polypeptide-specific Antigen (TPS) has been demonstrated to be useful in diagnosis and monitoring of adult epithelial tumors. So far, no data have been available on normal or pathologic TPS values in children. Therefore, the present study was designed to evaluate the normal values of TPS in childhood. Using a commercial enzyme linked immunosorbent assay (ELISA) kit, serum TPS was determined in 361 healthy children. Median (M) TPS was found to be 107 U/l at birth (n = 124). By the end of the first week, the value rose to M = 150 U/l (n = 68) and then continuously decreased with age (1 week–1 year, n = 45, M = 88 U/l; 1–7 years, n = 75, M = 51 U/l) until reaching the adult level (8–18 years, n = 49, M = 34 U/l). Additionaly, the serum TPS values of 45 mothers right after delivery (M = 161 U/l) were assessed, and there was no correlation to the marker levels determined in the cord blood of their children. The age-dependent distribution of serum TPS in healthy children must be taken into account in the clinical application of this tumor marker. Med. Pediatr. Oncol. 29:218–221, 1997.

Key words: tumor marker; intermediate filaments; cytokeratin 18; tissue polypeptide-specific antigen; normal value

INTRODUCTION

The tissue- and cell-specific expression of intermediate filaments (IF) are of particular use in tumor typing [1]. The most complex subgroup with 20 identified subtypes are the cytokeratins (CK) [2,3]. As an essential component of the cytoskeleton of epithelial cells, they are also believed to have a function in cell interaction [4]. Currently, tissue polypeptide antigen (TPA), tissue polypeptide-specific antigen (TPS), and Cyfra 21-1 [5] are used as cytokeratin-derived tumor markers.

TPA is a component of CK 8, 18, and 19 [6] and the assay is highly cross-reactive with keratin degradation products, resulting in a loss of specificity [7].

TPS has been identified by using a high-affinity monoclonal antibody against M3, one of 35 identified TPA epitopes, which constitutes the specificity related to cell proliferation [8]. TPS is identical with CK 18 and the distribution is quite different from TPA.

TPS measured in the cell-culture supernatant was found to correlate with cell number as well as DNA synthesis rate [9] and can be measured in serum using a commercially available enzyme linked immunosorbent assay (ELISA) kit. The value of this tumor marker was investigated in clinical trials on breast [10,11], ovary [12], prostate, renal [9,13], and colon [14] cancer.

Normal values for serum TPS in children have not been available so far; only TPA levels for healthy children have already been published [15]. In a preliminary report, we hypothesized that TPS may be of diagnostic value in the diagnosis, therapy, and follow-up of pediatric tumors [16]. The aim of the present study was to assess normal TPS values for all age groups of healthy children.

MATERIALS AND METHODS

The serum TPS values of 361 Caucasian children (202 males, 159 females) were evaluated. Except newborns and 1-week-old infants, all study subjects were admitted to our department for elective minor surgery (such as non-incarcerated inguinal hernia, umbilical hernia, hydrocele, and phimosis). All of the children included in this study were healthy as previously assessed by a pediatrician (because of planned general anesthesia). Serum TPS values in 124 newborns were determined from umbilical cord blood; complications of delivery and perinatal infections were excluded. In 14 of them we also

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measured the TPS value 1 week after birth. These 14 newborns were a representative sample of the whole population.

Homogeneous age distribution was achieved by allotting the subjects to different age groups: **1**) full-term newborns at birth (n = 124); **2**) 0–1 week (n = 68); **3**) 1 week–1 year (n = 45); **4**) 1–7 years (n = 75); and **5**) 8–18 years (n = 49).

Additionally, the serum TPS values of 45 mothers right after delivery were evaluated (prematurity, complications of pregnancy, and cesarean sections were excluded); in 20 cases we simultaneously obtained cord blood from their newborns.

For measurement, whole venous blood samples (without additives) obtained preoperatively from routine blood tests were centrifuged at 1,500 rpm for 10 min immediately after drawing and 500 μl of the supernatant was deep frozen at −70°C. Serum TPS values (U/l) were measured using the M3-Mab in an ELISA kit (BEKI Diagnostics, Bromma, Sweden). All serum samples were tested in duplicate according to the manufacturer's recommendations. The coefficients of variation between the assays were 3.6% and 8.7%, respectively, as determined by two control samples run in each assay. The control samples contained low and high concentrations of TPS.

Box plots were used to show TPS medians and quartiles of various age groups. All values are expressed as median (M).

Statistics

Since TPS values were not normally distributed in the various age groups, comparisons were performed by nonparametric unpaired or paired tests, corrected for ties and for multiple comparisons when necessary (refer to Fig. 1 legend for details). Pearson's and Spearman's rank correlation coefficients were calculated for quantitative variables. The SPSS statistical software (SPSS for Windows 6.0.1, SPSS Inc., Chicago, IL) was used.

RESULTS

Median TPS values (M) and IQR (interquartile range: Q1–Q3) of various age groups in healthy children are shown in Figure 1. The TPS value in infants at the end of the first week (n = 68), seemed to be significantly higher than in newborns (M = 150 U/l, $P \leq 0.003$). To further test for this hypothesis, we selected a sample of 14 neonates and measured TPS both in their cord blood at birth and in their serum at the end of the first week (see Fig. 1 inset). Thirteen of them exhibited an increase of their initial serum value. This increase was again significant (n = 14, $P \leq 0.002$).

Children obviously exhibit an age-dependent distribution of serum TPS. Until the end of the first year, the median serum value decreases to 88 U/l (n = 45). A further decrease of serum TPS levels could be observed in the following age groups: 1–7 years (M = 51 U/l, n = 75) and 8–18 years (M = 34 U/l, n = 49). There were no significant differences between males and females in either group.

The serum TPS values of mothers right after delivery (n = 45, M = 161, IQR 107–223 U/l) did not correlate with age of mothers or duration of pregnancy.

The serum TPS in newborns did not correlate with birth weight (M = 3,300 g). TPS seems to correlate with gestational age (range: 36–42 weeks), but this relation fell short of statistical significance ($P \geq 0.05$).

The TPS value of mothers at the time of birth does not correlate with the cord blood TPS of their children (n = 20).

DISCUSSION

We have assessed TPS values in 361 healthy children and found that the marker concentration is not constant with age; rather, the highest serum concentration can be measured in 1-week-old infants and thereafter decreases and reaches adult levels at about 8 years. Therefore, we considered it appropriate to establish definite age groups (i.e., up to 1 year, 1–7 years, 8–18 years). These age groups proved to be significantly different from each other ($P \leq 0.001$).

Between 8 and 18 years, TPS values even seem to be lower than reported adult values (M = 43.9 U/l in 206 healthy subjects between 18 and 65 years; Björklund, personal communication).

The reasons for this age dependence remain unclear, but several studies [9,17,18] showed that TPS is a marker of proliferation activity, so that higher TPS values could be expected during growth. In analogy to this hypothesis, Marcon et al. [19] described an association between rising TPS serum concentrations in pregnant women with the increase of the fetus cell mass, independent of the fetus' actual weight.

We cannot explain why serum TPS exhibits a peak concentration at the end of the first week. Probably the immaturity of (TPS) degradation and excretory mechanisms during the first days of life is responsible. Adaptation of liver function plays a pivotal role in the postnatal period, and at present hepatic CK metabolism is being intensely studied.

In 45 mothers right after delivery, we found a mean serum level of 173 U/l, which is remarkably lower than the value previously reported by Inaba et al. [17] (mean = 824 U/l, n = 5). This discrepancy can be due to racial differences—which the authors deny—or to the use of a different technique (BEKI IRMA [immunoradiometric assay] kit). Marcon et al. [19], however, found an excellent correlation between the ELISA and IRMA methods in 42 cases.

Serum marker levels of mothers do *not* correlate with values measured in their offspring. Since placental trans-

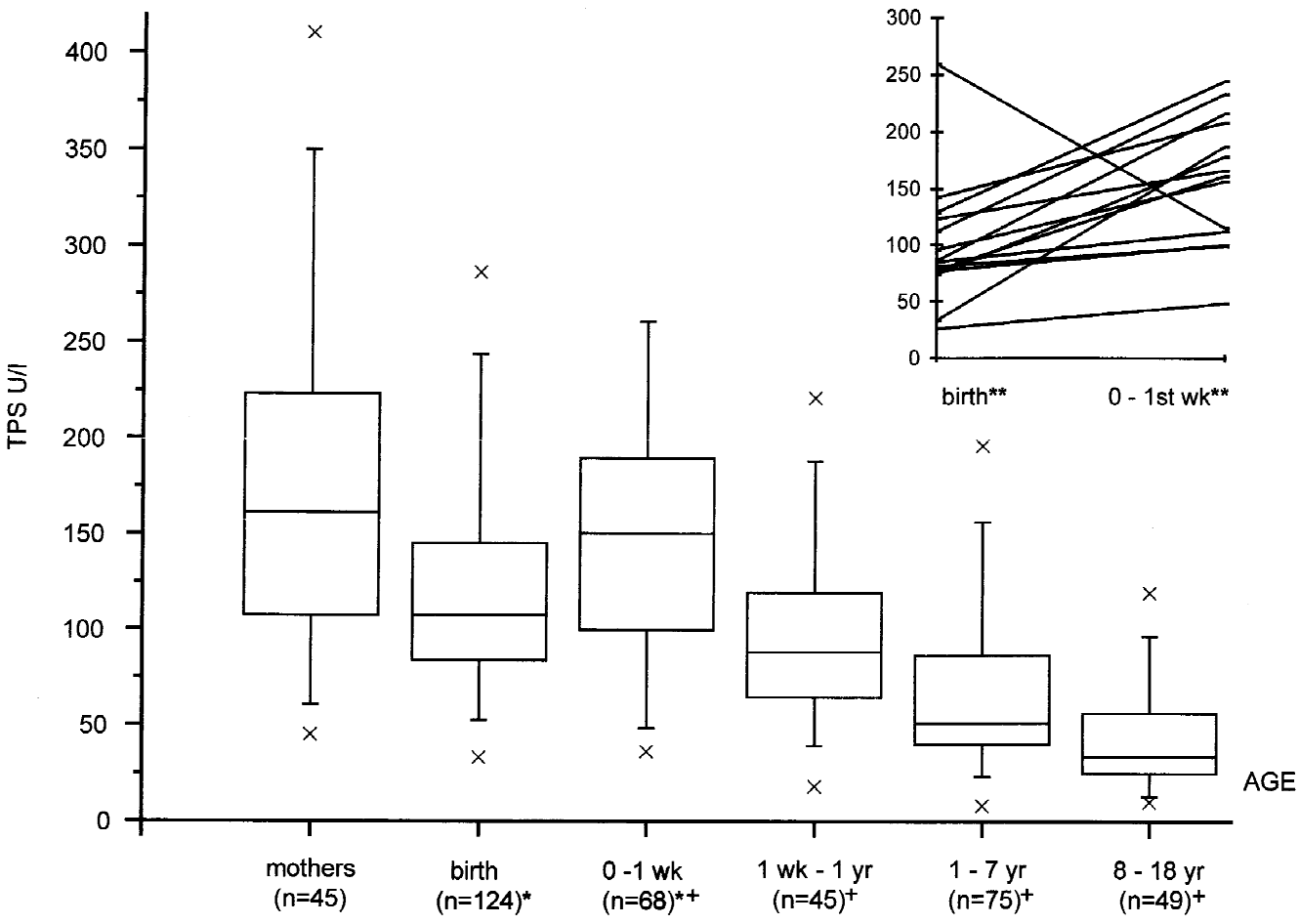

Fig. 1. Box and whiskers plot of serum TPS values in 45 mothers and 361 healthy children showing medians; 1st and 3rd quartiles (boxes), 5th–95th percentile range (whiskers), and minimum and maximum values (x) for the respective (age) groups. Note that there are significant differences between groups: $*P \leq 0.003$ (Mann-Whitney U-test) and $^{+}P \leq 0.001$ (Mann-Whitney U-test with Bonferroni correction for multiple comparisons). **Inset:** Consecutive measurements of serum TPS values in 14 selected cases at birth and after 1st week of life ($**P \leq 0.002$, Wilcoxon signed rank test). The 14 cases were a representative sample of the whole population (Locke's location compatibility test).

fer and metabolism of TPS are still a matter of speculation, we cannot offer a satisfactory explanation for this phenomenon.

Assessment of normal values is mandatory in the critical appraisal of a tumor marker. We hope to have contributed to this aim by measuring serum TPS in healthy children, especially since preliminary data [16] already indicate that TPS can be of some value in the clinical management of various pediatric malignancies.

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